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Review Article

HERBAL NOVEL DRUG DELIVERY SYSTEMS AND TRANSFERSOMES

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ABSTRACT

The strength of any herbal formulation depends on the delivery of phytoactives to an effective level. This limitation can be overcome by development of novel drug delivery systems, which ensure optimized drug delivery, enhanced bioavailability and better stability of phyto constituents for better therapeutic effects. Several novel herbal delivery systems have been successfully developed in recent years like liposomes, phytosomes, solid-lipid nanoparticles, ethosomes, microemulsions and various other vesicular systems. Transfersomes are vesicular drug delivery system having almost same structure like liposomes, but with better skin penetration properties to deliver the drugs at deeper skin tissues. Transfersomes are better drug delivery agents due to their ultradeformable structure.

Keywords: Herbal, Novel Drug Delivery, Transfersomes

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INTRODUCTION

From time immemorial it has been the endeavor of the physician and the apothecary to provide patients with the best possible forms of medicines so that recovery from disease is faster and complete. The drugs are delivered in a suitable formulation keeping in view the safety, efficacy and acceptability among other factors, and the formulation is usually known as dosage form or drug delivery system. With the progress in all spheres of science and technology, the dosage forms have evolved from simple mixtures and pills to highly sophisticated technology intensive drug delivery systems, which are known as Novel Drug Delivery Systems (NDDS) ¹.

Much research is being carried out on single herbs, poly-herbal formulations or herbo-mineral compounds, pharmaceutical products, combined treatments and disease specific therapies. However, the path remains uncertain in terms of standardization of products along with safety and efficacy for universal acceptance ².

Herbal drugs are becoming more popular in the modern world for their application to cure variety of diseases with less toxic effects ³.

The majority of biological active constituents in plants are water soluble like flavonoids, glycosides, tannins etc. These phytoconstituents are weakly absorbed, either owing to their giant molecular mass that can not be absorbed by inert transmission or owing to their reduced macromolecular solubility, a factor that severely restricts their capability to pass through the lipid rich natural membrane, following reduced bioavailability. In novel drug delivery technology, control of the distribution of drug is achieved by incorporating the drug in carrier system or by changing the drug behavior at the molecular level.

Some limitation of herbal extracts / plant actives are, instability at high pH, liver metabolism which has lead to drug level below therapeutic concentration in the blood, resulting in less or no therapeutic effects.

Other problems which are associated with herbal formulation are dose adjustment, maintenance of drug release pattern, which reduced efficacy of such system.

Advantage of Herbal Novel Drug Delivery System

Delivery of phytoactives through novel drug delivery systems has various advantages over conventional dosage forms.

- Incorporation of novel drug delivery technology to herbal of phytoactives minimizes the drug degradation or pre-systemic metabolism and other side effects by accumulation of drugs to the non-targeted areas and improves the ease of administration
- Novel drug delivery system is advantageous in delivering the herbal drug at predetermined rate and delivery of drug at the site of action, which minimizes the toxic effects with increased bioavailability of drugs.
- Incorporation of herbal drugs in the delivery system also aids to increase in solubility, enhanced stability, protection from toxicity, enhanced pharmacological activity, improved tissue macrophage distribution, sustained delivery and protection from physical and chemical degradation.
- Throughout the extraction method, the drug molecules within the herbal medication are exposed to oxidation, reduction or many other chemical reactive functional groups which cause degradation in phytoactive molecules. A nanocoating of those

drug molecules protects the active chemicals from degradations and thus enhance the stability and efficacy.

- It may help in increasing the efficacy and reducing the side effects of various herbal compounds and herbs.
- Some drugs have an optimum concentration range within which maximum benefit is derived, and concentrations above or below this range can be toxic or produce no therapeutic benefit, herbal novel drug delivery systems eliminates this limitation by delivering the correct amount of drug at predetermined controlled manner.

Types of herbal Novel Drug Delivery Systems

There are various novel drug delivery systems for herbal molecules are based on:

(a) Vascular delivery systems:

Transfersomes, Ethosomes, Liposomes, Phytosomes

(b) Nano drug delivery systems:

Micropallets, Microspheres, Nanoparticles

(c) Biphasic drug delivery systems:

Micro and Nano-emulsions

Table 1: Various Novel Drug Delivery Systems for Phytoactives.

Delivery System	Description	Ref
Invasome	Invasomes are the liposomal vesicles, which act as potential carriers with increased skin penetration. Invasomes have higher penetration rate through the skin as compared to liposomes and ethosomes.	4
Ethosomes	Ethosomes are noninvasive delivery carriers that enable drugs to reach the deep skin layers and/or the systemic circulation. These are soft, malleable vesicles tailored for enhanced delivery of active agents.	5
Pharmacosomes	Pharmacosomes are colloidal dispersions of drugs covalently bound to lipids, and may exist as ultrafine vesicular, micellar, or hexagonal aggregates, depending on the chemical structure of drug-lipid complex.	6
Vesosomes	A Vesosome is a more or less heterogeneous, aggregated, a large lipid bi-layer enclosing multiple, smaller liposomes that offers a second barrier of protection for interior components and can also serve as the anchor for active targeting components.	7
Sphingosomes	Sphingosomes is bilayer vesicles in which an aqueous volume is entirely enclosed by membrane lipid bilayer mainly composed of natural or synthetic sphingolipid. Sphingosomes showed higher stability, less in-vivo circulation time. Used to deliver chemotherapeutic agent and biological macromolecule.	8
Niosomes	Niosomes are non-ionic surfactant based vesicles that have a similar structure to that of phospholipid vesicles like liposomes. They have ability to increase the stability of entrapped drugs, improved bioavailability of poorly absorbed ingredients and enhanced skin penetration.	9
Nanoparticles	Solid lipid nanoparticles are composed of lipid matrix, which becomes solid at room temperature and also at the body temperature. The main features of solid lipid nanoparticles (SLNs) with regard to parenteral application are the excellent physical stability, protection of incorporated labile drugs from degradation.	10
Nano-emulsions	Micro-emulsion is also called nano-emulsion, and the sub-micro-emulsion is also called lipid emulsion. Emulsion drug delivery system is targeted or distributed well due to affinity to lymph Micro-emulsions are solutions containing nanometre-sized droplets of an immiscible liquid dispersed in an aqueous buffer.	11

Table 2: Various Vesicular Systems used for herbal drug delivery

Vesicular System	Advantages	Limitation
Liposomes	Biocompatible, Biodegradable	Stability issue, less penetration property
Niosomes	Improved stability due to use of non-ionic surfactants	Skin penetration is not much effective
Transfersomes	Ultradeformable with high stability, high penetration property, biodegradable, bio-compatible, incorporate low and high molecular weight drugs, penetrate deeper skin regions.	Difficulty in loading hydrophobic drugs
Phytosomes	Higher Bioavailability, enhanced capacity to cross the lipid rich biomembranes, better pharmacokinetic and therapeutic profile	leaching of the phytoconstituents
Ethosomes	Increased stability, increased skin permeability and inexpensive to formulate	Loss of product during transfer form organic to water media

TRANSFERSOMES

The name means “carrying bodies”. Transfersomes word is derived from the latin word ‘transferee’ which means ‘to carry across’ and the greek word “soma” which is used for a body. Transfersomes are a type of liposomes¹².

The basic structure of transfersomes is like classic liposomes, still it has some differences from liposomes by soft nature, ultra-deformable properties, and better adjustable nature of system membrane.

An important property of transfersomes is its ability to bind with skin moisture and retain water. Transfersomes contains high amount of hydrophilic molecules to avoid dehydration.

The first generation of Transfersomes includes vesicles, composed of phospholipids with edge activators. The second generation of transfersomes is composed by a combination of a bilayer component (e.g. phosphatidylcholine) and more than one amphipathic membrane destabilizing component. The third generation of transfersomes is constituted by amphipathic non-phospholipidic bilayers, but unlike the first and second generation, the surfactant is replaced by water soluble modulator compounds (e.g. organic ions) that have the same efficacy¹³.

When the transfersomes applied to the skin they move deeper towards the water containing strata to acquire their hydration. Skin barrier penetration of transfersomes is due to reversible bilayer ultradeformation, but without losing its integrity¹⁴.

It was claimed that the presence of water gradient exert a force in the order of 10^{-9} N per vesicle of a radius of approximately 60nm. As a result of this force the deformable vesicles can be squeezed through lipid bilayer. A low interracial tension makes them more deformable¹⁵.

Transport driving force magnitude plays an important role on the penetration, as

Flow = area X force X permeability

The mechanism of transfersomes penetration can be explain in steps as initial interaction between hydrophilic lipid residue and proximal water, from there the polar lipid attracts water molecules, which leads induced hydration, the vesicle moves toward the site of more water concentration. A trans-epidermal osmotic gradient develops, leads to penetration of transfersomes across skin¹⁶.

Another possible explanation for the fact that transfersomes are able to deliver molecules under non-occluded conditions could be that, as a result of water evaporation from the applied sodium-cholate phospholipid aggregate system, concentrated micellar system of cholate or collate-phospholipid or both generated. The micelles may further delipidize the stratum corneum creating small pores through which drug penetrates.

Methods of Transfersomes Preparation

Various chemicals are used in formulation of transfersomes, having various roles i.e. phospholipids, surfactants, dyes, alcohol, buffering agents etc.

Table 3: Various ingredients and their role for synthesis of Transfersomes

Ingredients	Chemical	Role
Phospholipids	Phosphatidyl choline, soya Phosphatidyl choline, dipalmitoyl phosphatidyl choline	Formation of vesicles
Surface active agents (edge activators)	Sodium Cholate, Sodium deoxycholate, tween 80, span 80	Flexibility improvement
Primary alcohol	Ethanol, Methanol	Solvents
Buffering agents	pH 6.4, phosphate Saline	Hydration medium

Thin film hydration method

In this method, phospholipids and surfactants are dissolved in suitable organic medium, such as chloroform-methanol and prepared a thin film, using rotary evaporator. The organic solvent evaporated at above the lipid transition temperature at 50°C.

Saline phosphate buffer of pH 6.4 is added to hydrate stack of film and subsequently kept for rotation at 60rpm for 1hr. kept at room temperature for 2 hrs, till the swelling of vesicle is completed. Afterwards to achieve desired size, the dispersion is sonicated¹⁷.

Modified Hand Shaking Lipid Film Hydration Method

Phytoconstituent, phosphatidyl choline (lecithin) and other additives, such as edge activators (sodium cholate) are dissolved in chloroform-ethanol mixture (1:1). The mixture of organic solvent is removed by evaporation by using rotary evaporator or by hand shaking at approximate 43°C.

A stack of thin film of lipid is formed over the inner wall of flask. The obtained stack on inner wall is kept overnight under vacuumed for complete removal of traces of solvents. The phosphate buffer (pH 4) is added to rehydrate the film with hand shaking for 15 min. at mentioned temperature. The vesicle of desired size can be obtained by sonication¹⁷.

Optimization factors of transfersomes formulation

The numbers of process variables are responsible to obtain the optimized formulation of transfersomes. There are various factors that are responsible for the same are:

1. pH

The pH evaluated by potentiometry affects the molecule ionization and consequently its interaction and entrapment efficiency. The pH level should be suitable for achieving a balance between formulation properties and biological applications, including the administration route^{18, 19}.

2. Effect of phosphatidyle choline, edge activator ratio-

These should be an optimized ratio of phosphatidylecholine and edge activator, because this affects greatly the entrapment efficiency. Higher concentration of edge activators may decrease entrapment efficiency. Upon incorporation of edge activators in low concentration, growth in vesicle size occurred²⁰, whereas further increase in the content of edge activator may have led to pore formation in the bilayer and reduced penetration.

3. Effect of type of edge activator-

Deformability and entrapment efficiency of transfersomes is affected by the type of edge activators, which could be interpreted by difference in their chemical structure. One way to explain this effect is to consider the HLB of edge activators. In one finding, tween 80-3 showed higher deformability. This could be attributed to the highly flexible and non-bulky

hydrocarbon chain of tween-80. Sodium deoxycholate (SDC-3) and Sodium cholate (SC-3) has lower deformability than tween-80-3 due to thin steroid like structure, which are bulkier than hydrocarbon chains of tween 80. This could be a result of their high hydrophobicity, which reduced the formation of transient hydrophilic holes, hence, minimizing the amphiphilic property of the bilayer responsible for membrane fluidity²¹.

4. Effect of Total lipid concentration-

Various research findings indicated that the fraction of lipid taking part in encapsulation was reduced upon increasing the total lipid concentration. Total lipid concentration used for formulation affects entrapment efficiency²².

Edge activators influenced the vesicular morphology, entrapment efficiency, zeta potential, drug release, deformability, skin permeation, and skin deposition.

Evaluation Parameters

Different parameters are used for characterization and optimization of transfersomes:

(A) **Entrapment efficiency**- the entrapment efficiency is the amount of drug entrapped in the formulation. This is determined by separating the entrapped drug in vesicles by using various techniques i.e. mini-column centrifugation. After that the vesicles were disrupted using 0.1% triton X-100 or 50% n-propanol.

The entrapment efficiency is calculated by using this formula

$$\text{Entrapment efficiency} = \frac{\text{amount of drug entrapped}}{\text{total amount of drug added}} \times 100$$

(B) **Diameter of Vesicle**- Photon correlation spectroscopy or dynamic light scattering method is used to determine vesicle diameter. Distilled water is used for the preparation of sample and filtered through a membrane filter (0.2 mm) and filtered saline used to dilute up to certain dilution ratio to measure the size of vesicles.

(C) **Degree of deformability or permeability measurement**- this is an important parameter for characterization as this greatly affects the permeation of formulation. This study is done by passing the preparation through a large number of pores of known size (micro porous filters of 50-400 nm)²³.

Particle size and size distribution are noted after each pass by dynamic light scattering (DLS) measurements.

$$D = J / (rv / rp)$$

Where

D= degree of deformability

J = amount of suspension extruded during 5 min.

rv = size of vesicle

rp = pore size of barrier

- (D) **Measurement of turbidity:** Nephelometer is used to measure turbidity sample in aqueous solution.
- (E) **Surface charge and charge density:** Zeta sizer is used to measure the surface charge and charge density on transfersomes.
- (F) **Number of vesicles per cubic mm-** Unsonicated transfersomes formulation, diluted 5 times with 0.9% sodium chloride solution. Hemocytometer with optical microscope are used for this study.

The transfersomes in 80 small squares are counted and calculated using formula;

$$(\text{Total number of transfersomes}) \text{ mm}^3 =$$

$$\frac{\text{Total number of vesicle counted} \times \text{dilution factor} \times 4000}{\text{Total number of square counted}}$$

- (G) **Vesicle Morphology-** vesicle diameter can be determined using photon correlation spectroscopy (PCS) or Dynamic Light Scattering (DLS). Samples were prepared in distilled water, filtered through a 0.2 mm membrane filter and dilutes with filtered saline and then size measurement can be done by using PCS or DLS.

Transfersomes vesicles can be visualized by Transmission electron microscopy (TEM) or phase contrast microscope. Mean size is measured by DLS and TEM, structural changes are observed by TEM ²⁴.

APPLICATION OF TRANSFERSOMES

Transfersomes have a wide variety of applications in the targeting of drug and other molecules through the transdermal route.

1. Delivery of Proteins-

It is very difficult to transport big and large biogenic molecules such as body proteins and peptides into body. When administered through oral route, such molecule shows degradation in gastrointestinal tract ²⁵.

Transfersomes are the best suitable approach for the delivery of all kinds of proteins into body. It is observed that bioavailability of the molecules delivered by transfersomes are similar to the drug administered by subcutaneous injections ^{26, 27}.

The protein preparation e.g. bovine serum albumin (immunogenic adjuvant) applied repeatedly in the preparation of transfersomes through epi-cutaneous route, showed strong immunogenic response.

Transfersomes has been shown to penetrate the skin intact, to present the antigen to dendritic cells and elicit an antigen specific immune response ²⁸.

Gap junction proteins loaded in transfersomes elicit antigen specific antibody titer that was equivalent to subcutaneous route ²⁹.

Plasmid DNA encoding Hepatitis-B surface antigen (HBs-Ag)- loaded cationic transfersomes are also utilized for topical immunization, and showed significantly higher HBs-Ag antibody titer and cytokinins level ³⁰.

2. Delivery of Anti-cancer drugs

Transfersomes are used as carrier for delivery of anti-cancer drugs; they are suitable specially for treating skin cancers. Transfersomes loaded with methotrexate was tried for treatment of skin cancer. Tamoxifen (TAM) anti breast cancer agent is carried through skin most efficiently by means of transfersomes and accelerate the growth of murine uteri, where it act as an anti-oestrogen, even at low dose as 0.1-0.2 mg/kg/day ³¹.

3. Delivery of Interferon-

Transfersomes loaded with immunomodulators, Interferon- α and interleukines-2 (IL-2) are successfully synthesized and observed that both the molecules retained their biological activity and could be efficiently encapsulated in carrier ³².

4. Insulin Delivery-

Orally applied polypeptidic or proteinaceous drugs are digested in the gastro-intestinal tract, by and large, and thus are therapeutically nearly inactive. Transfersomes can transport their associated drugs, including the epicutaneously applied insulin, into the body spontaneously. This happens in spite of the fact that insulin is normally prevented from crossing the skin by its high molecular weight of 5808 Da. The self-regulating membrane deformability of transfersomes is closely related to the corresponding vesicles self-reparation capability, the latter being essential for the transfersome stability and practical usefulness. Insulin is inferred to be transported into the body between the intact skin cells with a bio-efficiency of at least 50% of the subcutaneous dose action ³³.

5. Delivery of various therapeutic agents

Table 2: Various Drugs used with Transfersomal drug delivery system

Drug	Category	Therapeutic activity	Ref
Dexamethasone	Corticosteroid Drug	Anti-edema activity	34
Diclofenac	NSAID agent	Formulation optimization	35
Tacrolimus	Immunosuppressive	Atopic dermatitis	36
Pentoxifyllin	Xanthine Derivative	Chronic occlusive arterial disease.	37
Eprosartan Mesylate	Angiotensin receptor blockers (ARBs)	Management of Hypertension	38
Ciprofloxacin	Quinolone Antibiotic	Treatment of otitis media	39
Timolol maleate	Nonselective β -adrenergic blocker	Management of Hypertension	40
Ketoconazole	Azole antifungal	Antimicrobial activity	41

Diclofenac diethylamine, Curcumin	NSAID and natural Phenol curcuminoid	Analgesic and Anti-inflammatory	42
Itraconazole	Antifungal triazoles	Formulation Optimization	43
Paromomycin sulfate	Antibiotic	cutaneous leishmaniasis	44
risperidone	antipsychotics	Formulation Optimization	45
Minoxidil and caffeine	Antihypertensive vasodilator	Treatment of alopecia	46
Raloxifene Hydrochloride	Selective oestrogen receptor modulator (SERM)	Treatment of osteoporosis	47
sinomenine hydrochloride	Alkaloid	Formulation optimization Treatment of Rheumatism	48
Embelin	Benzoquinone derivative	Treatment of Cancer	49
Indinavir sulfate	Protease inhibitor used as a component of highly active antiretroviral therapy to treat HIV/AIDS	Treatment against AIDS with other protease inhibitors, nucleoside analogues or reverse transcriptase inhibitors	50
Stavudine	Reverse transcriptase inhibitors	Prevention and treatment of HIV/AIDS	51

CONCLUSION

Transfersomes has several advantages of transdermal drug delivery system. Transfersomes can entrap and deliver small and large molecules effectively through skin. Their ultradeformable property makes them to overcome the skin permeation difficulty, as they squeeze themselves to cross the skin layer barriers. But for

optimized formulation the most critical factor is correct ratio of edge activators and phospholipids, which governs the flexibility, vesicle layers integrity, and entrapment efficiency and stability of the formulation. Transfersomes has explored for delivery of macro and micro molecules, but lots of phytoconstituents are there to explore the potential role of this delivery system.

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